



**STUDIES ON OPTIMUM FEEDING RATE, ENERGY  
AND PROTEIN MAINTENANCE REQUIREMENTS  
OF A SELECTED CULTIVABLE CATFISH SPECIES**

**DISSERTATION**

**SUBMITTED IN PARTIAL FULFILMENT OF THE REQUIREMENTS  
FOR THE AWARD OF THE DEGREE OF**

**Master of Philosophy**

**IN**

**ZOOLOGY**

**BY**

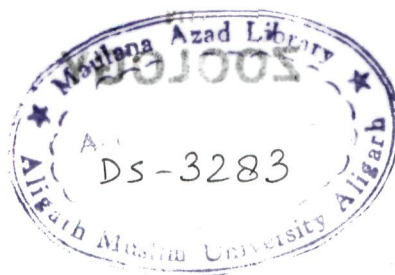
**HINA ALAM**

**FISH NUTRITION RESEARCH LABORATORY  
DEPARTMENT OF ZOOLOGY  
ALIGARH MUSLIM UNIVERSITY  
ALIGARH (INDIA)**

**2001**



DS3283





*A.K. Jafri*

M.Sc., Ph.D, F.N.A.Sc.

*Professor*

I certify that the work entitled “**Studies on optimum feeding rate, energy and protein maintenance requirements of a selected cultivable catfish species**” has been completed under my supervision by **Ms. Hina Alam**. The work is original and independently pursued by the candidate. It embodies some interesting observations contributing to the existing knowledge on the subject.

I permit the candidate to submit the work for the award of degree of **Master of Philosophy in Zoology** of the **Aligarh Muslim University, Aligarh, India**.

  
*A.K. Jafri*

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Fisheries Laboratory, Department of Zoology, Aligarh Muslim University,  
Aligarh, 202002, India. Tel (office) 91-571-400921-Extn. 301; (Res.) 91-571-401061;  
Fax: 91-571-400528; Email address: ahmadkjafri@yahoo.com

DEDICATED  
TO MY  
*PARENTS & TEACHER*

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## **ACKNOWLEDGEMENT**

It is my foremost privilege to acknowledge, with a deep sense of gratitude and humble submission, the able guidance, enthusiastic support, and never failing encouragement extended to me by my supervisor, Professor A.K. Jafri, Chairman, Department of Zoology, Aligarh Muslim University, Aligarh.

I am extremely thankful to my teacher, Dr. Mukhtar A. Khan, and my laboratory colleagues, Dr. Shabana Firdaus, Dr. Nazura Usmani, Ms. Rana Samad, Mr. Afzal, Mr. Imtiaz, Ms. Shabihul for their cooperation and help throughout the course of my study. Thanks are due to Mr. Nasiruddin for the help whenever sought for.

I extend my special thanks to my friends Nikhat and Farah for their cooperation and encouragement.

Last but not the least, I owe a lot to my loving parents, whose dedication, support and continuous encouragement enabled me to complete the task.

**HINA ALAM**

## **GENERAL INTRODUCTION**

Fish is a source of high quality animal protein. The increasing demand for high quality, low-cost protein has attracted global attention towards aquatic resources which can be exploited to the maximum through intensification of culture practices. The unpredictable pattern and decline in capture fisheries has further necessitated adequate attention to produce more through aquaculture.

In fish culture, semi-intensive and intensive practices are employed to get maximum yield in minimum time and available resources. Feed constitutes the major operational cost in such culture operations. It is, therefore, important that food provided to the cultured species should not only be cost-effective but also nutritionally balanced, and the species cultivated should be fast growing and disease resistant. In order to develop cost-effective and nutritionally balanced diet for a particular fish species, it is necessary to work out the nutrient requirements of the species concerned and formulate the feed accordingly. Although feeds have been developed for several cultivable fish species, intensification of culture for a large number of fish species, is still hampered by non-availability of practical diets matching to the nutrient requirements of the concerned species.

Many studies on fish nutrition in the past remained directed towards determining the nutrient requirements of cultivable fish species (Cowey and Sargent, 1972, 1979; Castell, 1979; Jauncey, 1982; NAS-NRC, 1983, 1993; Cho *et al.*, 1985; Wilson and Halver, 1986; Hepher, 1988; De Silva, 1989; Halver, 1989; Tacon, 1990; Arai, 1991; Boonyaratpalin, 1991; Hardy, 1991; Helland *et al.*, 1991; Kissil, 1991; Luquet, 1991; Satoh, 1991; Shimeno, 1991; Wilson, 1991; De Silva and Anderson, 1995; and Kaushik, 1995).

In India, both carps (exotic and indigenous) and catfishes are cultured in fresh waters. Among the catfishes, singhi, *Heteropneustes fossilis* and magur, *Clarias batrachus* have always been in great demand, fetching high price. Lately, other species of genus *Clarias*, namely, *Clarias gariepinus*, locally called as Thai magur has been clandestinely introduced in Indian waters from Thailand through Bangladesh, and is now being cultured by many fish farmers in the country. The fish is preferred over the native varieties because of its high yield. The other attributes that make the fish a suitable candidate species for aquaculture are high fecundity, survival rate, disease resistance, ability to accept wide range of food items, wide tolerance range to various environmental extremes and fairly good market value. The aquaculture potential of *C. gariepinus* has been discussed by several workers (Huisman and Richter, 1987). The fish has now become the center of active research in many countries across the world (Haylor, 1993).

Protein constitutes a major and most expensive component of formulated fish diet and is efficiently utilized by fish. It is a major constituent of fish tissue making up about 65-70% on dry weight basis. It is used for growth and repair and also readily catabolized by fish for various energy expenditures (basal metabolism and voluntary activity). A deficiency of non-protein energy sources in the diet results in greater utilization of protein for energy purposes, leading to retardation of growth and water quality deterioration due to excessive excretion of nitrogenous waste in the culture system (Beamish and Thomas, 1984). Therefore, while formulating fish diet other energy sources are invariably incorporated in the diet to ensure efficient utilization of protein for growth (Steffens, 1981; and Wilson and Halver, 1986). However, inclusion of excessive energy in the diet is also detrimental as fish feed



to meet their energy needs and stop feeding as soon as their energy requirements are met before the ingestion of sufficient amount of protein. In successful practical fish feed, therefore, protein is kept to a level which produces a good growth and feed conversion with minimum feed cost.

Dietary protein requirements in fish is influenced by a delicate balance of protein and energy in the diet (Wilson and Halver, 1986;and De Silva and Anderson, 1995). Optimization of E: P ratio in fish is widely accepted method to spare protein and increase growth at reduced cost. Many studies in the past were focused towards understanding the interaction between dietary energy and protein (Parazo, 1990; Jobling *et al.*, 1991; El Sayed and Tesheima, 1992; Hassan, 1993; Catacutan and Coloso, 1995; Hassan *et al.*, 1995; Elangovan and Shim, 1997; Hossain *et al.*, 1998; Jantrarotai *et al.*, 1998; Mc Googan, and Gatlin III, 1999;and Hernandez *et al.*, 2001).

Investigations have also been made on interactions between lipid or carbohydrate (as non protein energy source) and protein (Tabachek, 1986; De Silva *et al.*, 1991; Anwar and Jafri, 1995; Sakthivel and Baskaran, 1995; Steffens, 1996; Erfanullah and Jafri, 1998; Murty and Naik, 1999;and Chou *et al.*, 2001).

Maintaining proper energy level in the diet is also important from the viewpoint of its influence on body composition. Increasing energy density in the diet relative to protein results in accumulation of higher lipid in the fish (Zietler *et al.*, 1984;and Company *et al.*, 1999) which is undesirable and may reduce its marketability.

Protein and energy maintenance requirements have been worked out for a number of fish species by different workers (Gatlin *et al.*, 1986; Brown *et al.*, 1990; Hassan and Jafri, 1994;and Lupatsch *et al.*, 1998).

Knowledge of optimum feeding rate is equally essential for rearing a particular fish species irrespective of the fact whether a culture practice is intensive or semi-intensive. It minimizes the feed cost and checks water quality deterioration due to excessive feeding. The influence of feeding level on growth, feed efficiency and chemical composition of fish has been investigated for various fish species (Machiels and Henken, 1986; Hung *et al.*, 1993; Hassan and Jafri, 1994; Sampaio and Minillo, 1995; and Panda *et al.*, 1999). Feeding rate is also known to influence the requirement of major nutrients in fish (Tacon and Cowey, 1985; and Talbot, 1985). Several factors like size, and environmental conditions influence the feeding rate in fish (Hung and Lutes, 1987).

Although *C. gariepinus* has been extensively studied for various aspects of its nutrition, including feeding sequences, nutritional physiology and digestibility (Henken *et al.*, 1985, 1987; Degani *et al.*, 1989; Uys, 1989; Mybenka and Agua, 1990; Hoffman and Prinsloo, 1995; Awaiss and Kestemont, 1998; and Murty and Naik, 1999), only few workers have investigated the effects of feeding rate and dietary energy and protein levels on *C. gariepinus* (Machiels and Henken, 1985; and Henken *et al.*, 1986). Pantazis and Jauncey (1996) have also established the optimum dietary protein : energy ratio for the growth of this fish.

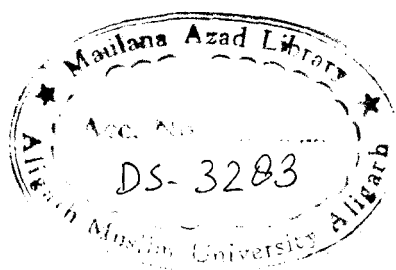
The present study deals with the optimum feeding rate, energy and protein maintenance requirements of *C. gariepinus* fingerling. A prerequisite to this study necessitated optimization of energy to protein ratio to be used in experimental diet for this species. This was achieved by examining the effects of dietary energy and protein levels, and their interaction on growth, utilization efficiencies and body composition of

the fish. The findings of these experiments have been outlined under two Chapters.

Chapter I describes the effects of dietary energy and protein levels, and their interaction on growth, utilization efficiencies and body composition of *C. gariepinus*.

Chapter II deals with the optimum feeding rate, energy and protein maintenance requirements of *C. gariepinus*.

The study may be useful for fish nutritionists as well as farmers, and find application in developing practical diets for the semi-intensive and intensive culture of this fish in Indian conditions.



## **GENERAL METHODOLOGY**

### ***I. Source of fish stock / acclimation***

Fingerlings of *Clarias gariepinus* were obtained from a fish farm in Rampur district, Uttar Pradesh. These were transported to the Research Station in oxygen filled polythene bags, given a prophylactic dip in KMnO<sub>4</sub> solution (1:3000) and stocked in flow-through (1-1.5L/min) outdoor cement cisterns (1x 1 x 1m) for a week. During this period fish were fed to satiation on minced meat twice daily at 0800 and 1600h. After a week, desired number of fish were taken out and acclimated on casein-gelatin based semi-purified diet (H-440) for two weeks in high density polyvinyl circular tanks ( water volume,70L) fitted with flow-through system (1L/min).

### ***II. Preparation of experimental diets***

Casein-gelatin based semi-purified diets were used for different experiments. Calculated quantities of dietary ingredients were weighed on a sensitive electronic balance (Precisa –120A). For preparation of diet, a weighed quantity of gelatin was mixed in a known quantity of water in stainless steel attachment of Hobart electric mixer with constant stirring, and heated to 80°C. The mixer bowl was then removed from heating, and weighed quantity of casein , dextrin, minerals and  $\alpha$ -cellulose were added to it, and the content blended in Hobart mixer while still in lukewarm state. This was followed by the addition of vitamin mix and oil (2:1, corn and cod liver oil). The mixture was again blended and in the end carboxymethyl cellulose was added to it. The prepared diet obtaining a bread dough like consistency, was poured into a teflon-coated pan and placed in a refrigerator to jell. The

prepared diet was in the form of moist cake, which was cut into small cubes and stored in refrigerator (-20°C) in sealed polythene packs until used. The mineral and vitamin premixes used (Table 1-2) were the same as given by Halver (1989).

### ***III. General experimental design / feeding trial***

Fish of desired size and number were sorted out from the acclimated stock and stocked in triplicate group in 70L polyvinyl circular flow-through (water volume 55L) troughs. The troughs were provided with ground water. The water exchange rate in each trough was maintained at 1-1.5L/min. Each morning faecal matter was siphoned off from the experimental troughs before feeding. Feeding level of fish and feeding schedule was chosen after carefully observing the dietary intake as well as feeding behaviour of fish. Fish were fed experimental diets in the form of moist cake six days a week. The moisture content of diet was estimated and ration level calculated as dry feed to wet fish weight. Mass weight of fish was taken weekly and amount of ration readjusted for subsequent feeding. On the day of weekly measurements no feed was offered to fish, when troughs were also thoroughly washed and rinsed with KMnO<sub>4</sub> solution. A record of daily dissolved oxygen and water temperature was maintained.

### ***IV. Proximate analysis***

Proximate analysis of carcass was made using standard techniques (AOAC, 1984). The analyses were carried out in triplicate runs.

#### ***(i) Estimation of Moisture***

For moisture estimation a weighed quantity of finely ground/homogenized sample was taken in a preweighed silica crucible

and placed in an oven ( $100^{\circ}\text{C}$ ) for 24 h. The crucible containing dried sample was directly transferred to a desiccator, allowed to cool and reweighed. This process was repeated till a constant weight is obtained. The loss in weight was expressed as per cent of moisture.

**(ii) Estimation of ash**

A known quantity of finely powdered sample was taken in a preweighed silica crucible and incinerated in a muffle furnace ( $600^{\circ}\text{C}$ ) for 2-3h till the sample became free of carbon. The crucible containing the incinerated sample was transferred to a desiccator, cooled and reweighed. The quantity of ash was calculated in percentage.

**(iii) Estimation of crude fat**

For estimating the crude fat, continuous soxhlet extraction technique was employed. Petroleum ether ( $40-60^{\circ}\text{C}$  B.P.) was used as solvent. A weighed quantity of finely ground sample was taken in Whatman fat extraction thimble, cotton plugged and introduced into the soxhlet apparatus. A clean dry soxhlet receiver flask was weighed and fitted to the soxhlet assembly on a water bath for extraction. Extraction was carried out for 10-12h. Thereafter, the flask was removed and kept in hot air oven ( $100^{\circ}\text{C}$ ) to evaporate the solvent traces. The flask was then cooled in a desiccator and then reweighed. The amount of fat extracted was expressed in percentage.

**(iv) Estimation of crude protein**

The technique employed for estimating the crude protein was based on a slight modification of Wong's micro-Kjeldahl method, as adopted by Jafri (1965). The principle involved digesting a known amount of sample in N-free sulphuric acid, in presence of potassium per-

sulphate used as catalyst, which converts the nitrogenous compounds to ammonium sulphate. This was then treated with Nessler's reagent. The colour developed due to the formation of a complex compound ( $\text{NHg}_2\text{I}$ ) was measured spectrophotometrically. The optical density obtained was read off against a standard calibration curve of  $(\text{NH}_4)_2\text{SO}_4$  for nitrogen estimation. To calculate the total crude protein in the sample, the amount of nitrogen was multiplied with the conventional protein factor (6.25).

0.1g dry powdered sample was taken in a Kjeldahl flask with 5 ml of N-free sulphuric acid (1:1), and .5 ml potassium persulphate added to it. The volume was raised to 3ml with distilled water. The solution was then nesslerized using Bock and Benedict's Nessler reagent, allowed to stand for 10 min before measuring the absorbance with a blank. The blank was prepared in the same manner using distilled water in place of aliquot. The amount of nitrogen was obtained by reading the optical density against the standard calibration curve (Fig. 1). The nitrogen value was multiplied with 6.25 to obtain the amount of crude protein. The spectrophotometric measurements were made on microprocessor- controlled split beam spectronic 1001 spectrophotometer (Milton Roy Company, USA).

#### ***(v) Estimation of gross energy***

Gross energy was calculated using fuel values 3.5, 4.5 and 8.5 kcal/g for carbohydrate, protein and lipid, respectively (Jauncey, 1982).

#### ***V. Assessment of growth and conversion efficiencies***

Calculation of growth parameters and conversion efficiencies were made according to standard definitions (Millikin, 1983; Tabachek, 1986; and Parazo, 1990).

$$\text{Increase in live weight (\%)} = \frac{W_2 - W_1}{W_1} \times 100$$

$$\text{Specific growth rate (\%)} = \frac{\log_e W_2 - \log_e W_1}{D} \times 100$$

Where,

$W_1$  = Initial mass weight (g)

$W_2$  = Final mass weight (g)

D = Duration of the feeding trial (days)

$$\text{Feed conversion ratio} = \frac{\text{Total feed intake (g)}}{\text{Live weight gain (g)}}$$

$$\text{Gross growth efficiency (\%)} = \frac{\text{Live weight gain (g)}}{\text{Total feed intake (g)}} \times 100$$

$$\text{Protein efficiency ratio} = \frac{\text{Live weight gain (g)}}{\text{Total protein intake (g)}}$$

$$\begin{aligned} \text{Protein productive value (\%)} &= 100 \times \left[ \frac{(\text{Final wet weight} \times \text{Final per cent body protein}) - (\text{Initial wet weight} \times \text{Initial per cent body protein})}{(\text{Amount of diet fed/No. of fish per trough}) \times \% \text{ crude protein in diet}} \right] \end{aligned}$$

Energy conversion efficiency (%)

$$= 100 \times \left[ \frac{(\text{Final wet weight} \times \text{Final body crude energy (kcal g}^{-1}\text{)}) - (\text{Initial wet weight} \times \text{Initial body crude energy (kcal g}^{-1}\text{)})}{(\text{Amount of diet fed/No. of fish per tank}) \times \text{Total energy in diet (kcal g}^{-1}\text{)}} \right]$$

## **VI. Statistical analysis**

The data were statistically analysed using standard methods (Snedecor and Cochran, 1967; and Sokal and Rohlf, 1981).



**Table 1. Composition of mineral mixture\***

Mineral	g/100g
Calcium biphosphate	13.48
Calcium lactate	32.40
Ferric citrate	2.97
Magnesium sulphate	13.70
Potassium phosphate (Dibasic)	23.86
Sodium biphosphate	8.72
Sodium chloride	4.35
Aluminium chloride .6H <sub>2</sub> O	0.015
Potassium iodide	0.015
Cuprous chloride	0.010
Manganous sulphate . H <sub>2</sub> O	0.080
Cobalt chloride .6H <sub>2</sub> O	0.100
Zinc sulphate. 7H <sub>2</sub> O	0.300

\*Halver (1989).

**Table 2. Composition of vitamin mixture\***

Vitamin	g/100g
Choline chloride	0.500
Inositol	0.200
Ascorbic acid	0.100
Niacin	0.075
Calcium Pantothenate	0.050
Riboflavin	0.020
Menadione	0.004
Pyridoxine-HCl	0.005
Thiamin-HCl	0.005
Folic acid	0.0015
Biotin	0.0005
$\alpha$ -tocopherol acetate	0.040
Vitamin B <sub>12</sub> (10 mg /500 ml H <sub>2</sub> O)	0.00001(0.5 ml)

\*Halver (1989).

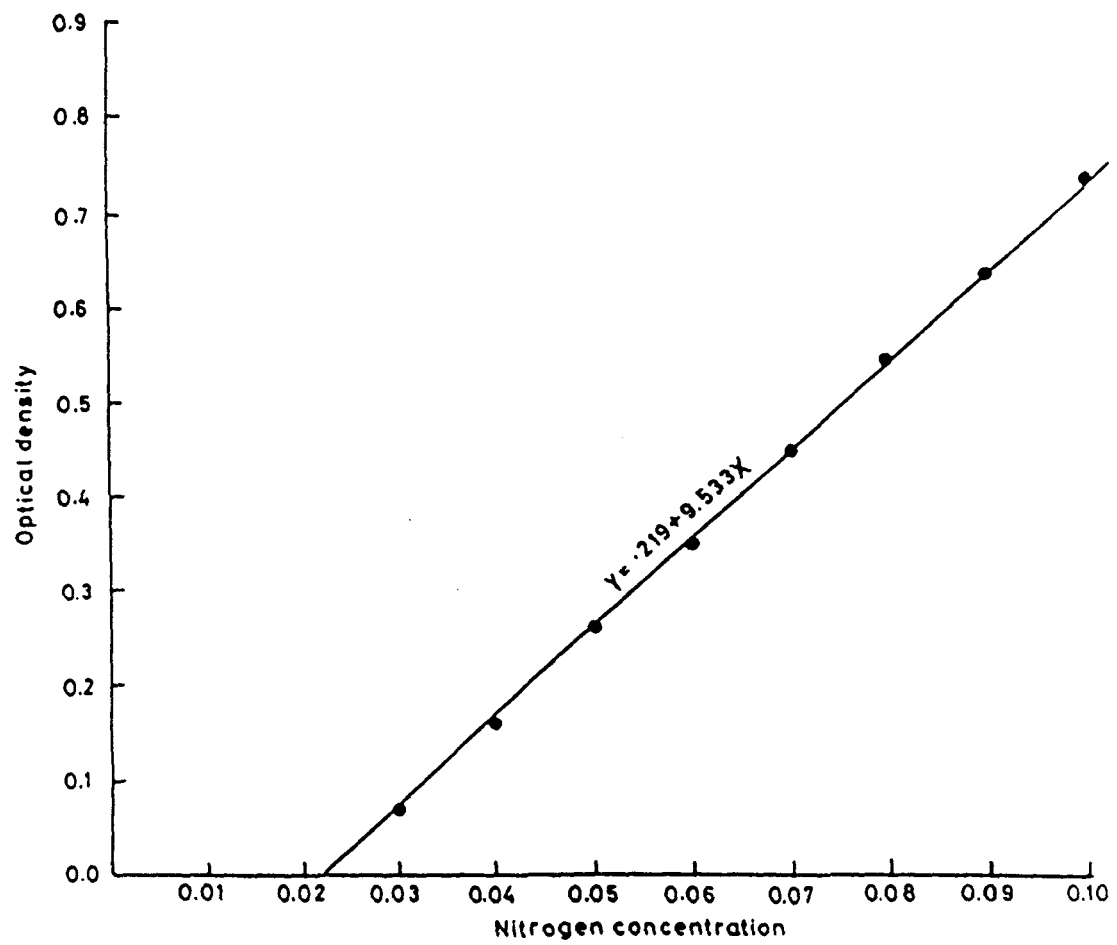


Fig. 1. Calibration curve of Nitrogen

## CHAPTER I

### EFFECTS OF DIETARY ENERGY AND PROTEIN LEVELS, AND THEIR INTERACTION ON GROWTH, UTILIZATION EFFICIENCIES AND BODY COMPOSITION OF CATFISH, *CLARIAS GARIEPINUS* (BURCHELL 1822).

#### INTRODUCTION

The determination of protein requirement is a prerequisite for the development of nutritionally balanced diet for the culture of a particular fish species. Higher levels of dietary protein, besides adding to the cost, also result in water quality deterioration due to excessive excretion of ammonia (Beamish and Thomas, 1984). The dietary protein requirement of fish is influenced, besides other factors, by a delicate balance of dietary energy to protein ratio. Provision of sufficient amount of non-protein energy in the diet results in greater utilization of protein for growth (Wilson, 1989; and De Silva and Anderson, 1995).

African catfish, *Clarias gariepinus* is regarded as important species for aquaculture. Although extensive work has been done on various aspects of nutrition of *C. gariepinus* (Henken *et al.*, 1985, 1987; Huisman and Richter, 1987; Degani *et al.*, 1989; Uys, 1989; Mybenka and Agua, 1990; Hoffman and Prinsloo, 1995; Awaiss and Kestemont, 1998; and Murty and Naik, 1999), little information exist on the effect of dietary energy and protein interaction on the growth and utilization efficiencies of this species (Machiels and Henken, 1985; Henken *et al.*, 1986; and Pantazis and Jauncey, 1996). The present study deals with

the effects of dietary energy and protein levels, and their interaction on growth and utilization efficiencies of *C. gariepinus* fingerling.

## **MATERIALS AND METHODS**

### ***Preparation of experimental diet***

Using semi-purified ingredients, experimental diets were formulated to contain three levels of protein (25, 40 and 45%), with each protein level containing two levels of energy (360.8 and 406.7 k cal/100g). Diet with 50% crude protein and 406.5 k cal/100g was used as control (Table 1). Dietary energy was calculated using physiological fuel values, 3.5, 4.5 and 8.5 kcal/g for carbohydrate, protein and fat, respectively (Jauncey, 1982). At each protein level, two different levels of lipids, 8 and 16%, were used. The amount of dextrin and  $\alpha$ -cellulose were altered to obtain the desired levels of energy in the diet. The method of preparation of diets has been described under General Methodology section.

### ***Feeding trial***

Source of fish, details of their acclimation, and general experimental design are described under General Methodology section. Young *C. gariepinus* ( $6.03 \pm 0.13$  cm;  $1.30 \pm 0.00$ g) were selected from among the acclimated fish and stocked in 70L polyvinyl circular troughs (water volume 55L), fitted with flow-through system (1L/min of ground water), in triplicate groups of 15 fish each, and fed control or one of the six experimental diets for 4 weeks. Average water temperature during the experimental period was  $26 \pm 1^\circ\text{C}$ .

Fish were fed till satiation at 0800 and 1600h. Uneaten feed, if any, was collected over a fine mesh sieve and dried overnight (100°C) for calculating the quantity of feed consumed.

The performance of diet, in terms of weight gain and nutrient utilization, was evaluated using standard definitions (page 10 ).

### ***Gross energy and proximate analysis***

20 fish were randomly taken out from the acclimated fish stock for the analysis of initial carcass composition, using standard techniques as given under General Methodology (page 6 ). At the end of feeding trial, 10 fish from each trough were again taken randomly, pooled and analysed for their final carcass composition. Gross energy was calculated using physiological fuel values (see page 9 ).

### ***Statistical analysis***

Statistical evaluation through one way analysis of variance (Snedecor and Cochran, 1967) and Duncan's multiple range test (Duncan, 1955) was done to test the difference between treatment means ( $P < 0.05$ ). Relationship between specific growth rate (%) vs. E/P ratio and dietary protein vs. weight gain (%) was established by linear regression analysis (Snedecor and Cochran, 1967).

## **RESULTS**

Results of feeding *C. gariepinus* with varying energy and protein ratios are given in Table 2 and 3. A significant influence of energy and protein was evident on fish growth in terms of per cent live weight gain. At each protein level, a general decline in per cent live weight gain was noticed with increasing dietary energy. Weight gain (%) was found to be

directly related (360.8 kcal/100g:  $r = 0.99$ ,  $n = 3$  and 406.7 kcal/100g:  $r = 0.96$ ,  $n = 4$  including control) to dietary protein levels (Fig.1). Diets with lower energy levels (360.8 kcal/100g) produced best growth. Maximum growth was observed at 45 % crude protein level with 360.5 kcal/100g energy.

Specific growth rate (SGR%) showed a negative linear correlation (Fig.2) with E/P ratios (360.5 kcal/100g :  $r = -0.99$ ,  $n = 3$  and 406.7 kcal/100g:  $r = -0.95$ ,  $n = 4$ , including the control diet).

The daily mean feed consumption (mg/fish day<sup>-1</sup>) was found to be influenced by both protein and energy levels of the diet. In general, at a given protein level feed consumption reduced with increase in dietary energy. Influence of dietary protein on feed consumption was positively linear (360.8 kcal/100:  $r = 0.99$ ,  $n = 3$  and 406.7 kcal/100g :  $r = 0.95$ ,  $n = 3$ ) at a given energy level. Correlation analysis between E/P ratio and feed consumption produced a negative linear effect ( $r = -0.84$ ,  $n = 6$ ). Fish fed 45% crude protein at lower energy density (360.5 kcal/100g) consumed maximum feed (539.4mg/fish day<sup>-1</sup>) while those fed with 25% crude protein and higher dietary energy (406.7 kcal /100g) consumed the minimum (375.8mg/fish day<sup>-1</sup>). The per cent live weight gain (Y) showed a positive correlation with daily mean feed consumption (X) ( $Y = -410.65 + 1.934X$  :  $r = 0.96$ ,  $n = 6$ ) (Fig 3).

Feed conversion ratio (FCR), protein efficiency ratio (PER), protein productive value (PPV) and energy conversion efficiency (ECE) were altered by both energy and protein contents of the diet. Best feed conversion was noticed in the group fed 45% crude protein and low energy. At a given energy level, FCR improved with decreasing E/P ratios (360.8 kcal/100g:  $r = 0.99$ ,  $n = 3$  and 406.7 kcal/100g :  $r = 0.99$ ,  $n = 4$ ). Poor FCR and lower values of PER, PPV and ECE were observed with increase in dietary energy content at a given protein

level. Both PER (360.8 kcal/100g:  $r = -0.99$ ,  $n = 3$  and 406.7 kcal/100g:  $r = -0.99$ ,  $n = 4$ ) and PPV (360.8 kcal/100g:  $r = -0.99$ ,  $n = 3$  and 406.7 kcal/100g:  $r = -0.96$ ,  $n = 4$ ) showed inverse relationship with dietary protein. An opposite trend was observed in ECE at both the energy levels (360.8 kcal/100g:  $r = 0.99$ ,  $n = 3$  and 406.7 kcal/100g:  $r = 0.98$ ,  $n = 4$ ) when isocaloric diets were compared. PPV and PER were maximum 30.71 and 3.62, respectively, at the lowest protein level (25%) with 366.6 kcal/100g diet, while maximum ECE (21.15%) was observed at 45% crude protein with 360.5 kcal/100g.

Moisture, crude protein, fat and ash in fish tissue were found to vary significantly among the various treatment groups (Table 3). At the end of feeding trial, fish in all the experimental groups showed higher percentages of lipid and lower percentages of moisture, protein and ash, compared to their initial values. A decrease in moisture, protein and ash was observed with increase in dietary energy.

Survival in each treatment was over 90%, and no specific pattern of mortality was noticeable to which treatment levels could be related.

## DISCUSSION

The results of the present study indicates that, as in many other fish species (Halver, 1989; and De Silva and Anderson, 1995), growth of *C. gariepinus* is also significantly influenced by the levels of energy and protein in the diet. Fish fed with 360.5 kcal/100g energy and 45% crude protein (E/P ratio 8.01) diet, produced maximum growth which was not significantly ( $P > 0.05$ ) different from the gains produced by the control diet with high energy (406.5 kcal/100g) and high protein (50% C.P.) having an E/P ratio of 8.13. When specific growth rate (SGR%) was



plotted against E/P ratio at both the energy levels (360.8 kcal/100g and 406.7 kcal/100g) also intersected at or near 8.01. A negative correlation existed between E/P ratio and SGR%, indicating that higher E/P ratios do not prove beneficial to fish in terms of growth. This fact is justified by a negative correlation observed between E/P ratio and the amount of feed consumed by the fish.

A positive correlation observed between the amount of feed consumed and gain in live weight per cent indicates that amount of feed consumed by the fish has a direct bearing on weight gain. Page and Andrews (1973) and Lovell (1979) made a similar observation on channel catfish. Hassan (1993) noted that in *C. batrachus* high digestible energy to protein ratio causes cessation of feeding before sufficient amount of protein is consumed by fish resulting in decreased fish growth. These observations strengthen the fact that feed consumption in fish is determined by energy density of the diet. A corollary to the above fact was also evident in the work of Jantrarotai *et al.* (1998) on hybrid *Clarias*, Li and Robinson (1999) on juvenile channel catfish and Hernandez *et al.* (2001) on sharpsnout seabream.

A general reduction in per cent gain in live weight with increase in dietary energy content at each protein level and a direct relationship with dietary protein content noted during the present study on *C. gariepinus*, conform to the findings of Hassan (1993) on *C. batrachus*, and McGoogan and Gatlin III (1999) on *S. ocellatus*. The observation that feed intake in *C. gariepinus* fingerling is significantly influenced by the levels of dietary energy and protein emphasizes that both energy and protein are important consideration while formulating the feed for fish.

The data indicates that, irrespective of the protein content of diet, feed consumption gets reduced with increase in dietary energy levels.

In *C. gariepinus* feed consumption was more pronounced in the groups fed low energy diets. Influence of protein and energy content on feed consumption has also been reported in other fishes (Tabachek, 1986; De Silva *et al.*, 1991; Hassan, 1993; Pullin *et al.*, 1996; Jantrarotai *et al.*, 1998; and Hernandez *et al.*, 2001).

Influence of both energy and protein contents on FCR, which reflects the overall performance of diet in terms of live weight gain, was also evident during the present study on *C. gariepinus*. Best FCR was obtained in fish groups fed diets containing 45% crude protein with 360.5 kcal/100g energy. Machiels and Henken (1985) and Uys (1989) also noted a dietary protein requirement of 40% or more for this species.

Protein utilization in fish is known to be influenced by quality of protein in the diet besides the levels of dietary energy (Steffens, 1981). In *C. gariepinus*, a negative correlation was evident between dietary protein and PER/PPV, with the highest values of both PER and PPV obtaining at lowest protein level (25%C.P.) Increase in dietary energy at each protein level, however, resulted in a reduction in both PER and PPV. Decreasing E/P ratio in isocaloric diets reflected a positive correlation with PER and PPV, indicating that more the dietary energy available  $\text{g}^{-1}$  protein, the greater is the utilization of protein for growth, this being measurable by PER and PPV. A negative correlation was observed between E/P ratio and ECE. Low ECE noted in the fish fed diet with high E/P ratio indicates that more energy is utilized to meet the energy demands of the body and little get deposited. Highest ECE was observed at 45% crude protein and 360.5 kcal/100g energy, and this was not significantly different from the value obtained with the control diet (50% crude protein and 406.5 kcal/100g energy), indicating that E/P ratio of 8.01 is adequate to meet the energy and protein

requirements of fingerling *C. gariepinus*. On the basis of weight gain and values of FCR, PER, PPV and ECE obtained by feeding fish with diet of varying E/P ration, it may be summarized that 45% crude protein diet with 360.5 kcal/100g energy is optimum for this fish.

The data on body composition of *C.gariepinus* reveals that proximate composition of fish is markedly influenced by dietary energy level. An increase in body lipid content observed in *C.gariepinus* with increase in dietary energy was also reported in other fish species (Zeitler *et al.*, 1984; Hassan, 1993; Catacutan and Coloso, 1995; and Hossain *et al.*<sup>a</sup>, 1998). Similarly, the inverse relationship noted between dietary energy (lipid) and body moisture, protein and ash of *C.gariepinus* was also seen in other fish species (Parazo, 1990; El Sayed and Tehsima, 1992; Hassan, 1993; and Panda *et al.*, 1999).

It is evident from the present study that to produce sufficient weight gain and better feed conversion, the diet of *C. gariepinus* should contain 45% crude protein and 360.5 kcal/100g of energy.

The dietary protein component can further be reduced at the same energy level to reduce feed cost. Reduced growth in fish fed low protein diet (25% C.P.) is compensated by relatively high protein retention. Therefore, it is suggested to make a compromise between growth rate and cost effectiveness of the diet while formulating feed for *C. gariepinus*.

## SUMMARY

Fingerling *C. gariepinus* were fed semi-purified diets, containing three levels of crude protein (25, 40 and 45%) and two levels of metabolizable energy (360.8 kcal /100g and 406.7 kcal/100g) at each protein level, over a period of 4-week feeding trial conducted in flow-

through troughs at  $26 \pm 1^{\circ}\text{C}$ , for evaluating the effect of dietary energy and protein on growth, utilization efficiencies and proximate composition of fish. Growth of fish, in terms of live weight gain (%), increased with increasing dietary protein at both the energy levels and at each protein level. Growth rate reduced with increase in energy density of the diet. SGR (%) exhibited a linear negative correlation with E/P ratio within isocaloric diets. Mean feed consumption, FCR, PER, PPV and ECE were influenced both by dietary energy and protein level in the diets within isocaloric diets. PER and PPV exhibited a positive relation, whereas FCR and ECE showed an increase correlation with E/P ratios. Carcass composition was influenced by both dietary energy and protein.

The study indicates that for optimum growth and better feed conversion practical diets for *C.gariepinus* fingerlings should contain 45% crude protein and 360.5 kcal/100g energy.

**Table 1. Ingredient and proximate composition of experimental diet**

Ingredient (g/100g, as fed)	Diets					
	Control	I	II	III	IV	V
Casein (Vitamin free; 84% C.P.)*	44.64	22.32	22.32	35.71	35.71	40.17
Gelatin (87.6% C.P.)*	14.26	7.13	7.13	11.41	11.41	12.84
Dextrin	13.00	53.16	47.16	30.66	24.66	25.71
Corn oil	10.64	5.34	10.64	5.34	10.64	5.34
Cod liver oil	5.33	2.66	5.33	2.66	5.33	2.66
Vitamin mix.	1.00	1.00	1.00	1.00	1.00	1.00
Mineral mix.	3.00	3.00	3.00	3.00	3.00	3.00
α-Cellulose	6.10	3.39	1.39	8.22	6.22	7.28
Carboxy methyl cellulose	2.00	2.00	2.00	2.00	2.00	2.00
<b>Proximate composition % (calculated)</b>						
Crude protein	50.00	25.00	25.00	40.00	40.00	45.00
Crude fat	16.00	8.00	16.00	8.00	16.00	8.00
Carbohydrate	13.00	53.00	47.00	31.00	25.00	26.00
Metabolizable energy (kcal/100g, as fed)	406.50	66.56	413.00	355.31	402.31	360.50
Carbohydrate calories (%)	11.19	50.75	39.96	30.20	21.45	25.24
Lipid calories (%)	33.45	18.55	32.92	19.13	33.80	18.86
Energy/Protein ratio (kcal/g protein)	8.13	14.66	16.52	8.88	10.05	8.01
						45.00
						16.00
						19.00
						405.00
						16.41
						33.58
						9.00

\*Loba Chemie, India.

**Table 2. Results of feeding *C. gartepinus*, experimental diets with varying E/P ratio**

	Diets						
	Control	I	II	III	IV	V	VI
Initial individual wet weight (g)	1.30 ± 0.01	1.30 ± 0.01	1.30 ± 0.00	1.32 ± 0.01	1.29 ± 0.01	1.30 ± 0.01	1.32 ± 0.01
Final individual wet weight (g)	9.77 ± 0.10	5.63 ± 0.05	4.75 ± 0.03	8.53 ± 0.31	6.90 ± 0.18	9.55 ± 0.07	7.83 ± 0.06
Percent gain in live weight	656.01 <sup>a</sup>	333.46 <sup>e</sup>	264.93 <sup>f</sup>	548.47 <sup>b</sup>	437.68 <sup>d</sup>	638.53 <sup>a</sup>	493.42 <sup>c</sup>
Specific growth rate (SGR%)	7.22 <sup>a</sup> ± 0.08	5.22 <sup>e</sup> ± 0.10	4.63 <sup>f</sup> ± 0.03	6.66 <sup>b</sup> ± 0.16	6.00 <sup>d</sup> ± 0.08	7.15 <sup>a</sup> ± 0.02	6.34 <sup>c</sup> ± 0.01
Feed consumed (mg/fish day <sup>-1</sup> )	536.64 <sup>a</sup> ± 1.53	384.89 <sup>cd</sup> ± 6.25	375.81 <sup>d</sup> ± 0.70	514.86 <sup>a</sup> ± 21.97	411.25 <sup>c</sup> ± 0.85	539.44 <sup>a</sup> ± 7.72	451.94 <sup>b</sup> ± 5.08
Feed conversion ratio (FCR%)	0.81 <sup>d</sup> ± 0.01	1.11 <sup>b</sup> ± 0.00	1.27 <sup>a</sup> ± 0.01	0.90 <sup>c</sup> ± 0.01	0.94 <sup>c</sup> ± 0.03	0.84 <sup>d</sup> ± 0.01	0.90 <sup>c</sup> ± 0.01
Protein efficiency ratio (PER%)	2.47 <sup>d</sup> ± 0.02	3.62 <sup>a</sup> ± 0.01	3.17 <sup>b</sup> ± 0.04	2.78 <sup>c</sup> ± 0.02	2.70 <sup>c</sup> ± 0.08	2.65 <sup>c</sup> ± 0.03	2.50 <sup>d</sup> ± 0.04
Protein productive value(PPV%)	20.24 <sup>e</sup> ± 0.12	30.71 <sup>a</sup> ± 0.29	24.52 <sup>b</sup> ± 0.20	24.08 <sup>b</sup> ± 0.27	23.00 <sup>bc</sup> ± 0.51	22.37 <sup>cd</sup> ± 0.41	20.90 <sup>de</sup> ± 0.97
Energy conversion efficiency (ECE%)	21.10 <sup>a</sup> ± 0.38	16.27 <sup>c</sup> ± 0.13	12.67 <sup>d</sup> ± 0.05	20.63 <sup>ab</sup> ± 0.13	19.53 <sup>b</sup> ± 0.42	21.15 <sup>a</sup> ± 0.43	20.27 <sup>ab</sup> ± 0.83
Percent Survival	100	100	100	100	93	100	100

Results are mean ± SE of triplicate fish groups  
Values in each row with similar superscript are insignificantly different (P>0.05)

**Table 3. Body composition of *C. gariepinus* fed experimental diets**

Diet	Energy (kcal/100g)	Moisture (g/100g, wet weight)	g/100g, dry matter		
			Protein	Fat	Ash
Initial		80.91 ± 0.12	69.36 ± 0.32	10.00 ± 0.00	8.52 ± 0.01
<b>25% crude protein</b>					
Diet I	366.56	76.12 <sup>a</sup> ± 0.13	65.56 <sup>a</sup> ± 0.16	19.99 <sup>d</sup> ± 0.11	7.20 <sup>ab</sup> ± 0.12
Diet II	413.00	76.04 <sup>a</sup> ± 0.30	63.80 <sup>b</sup> ± 0.12	24.37 <sup>b</sup> ± 0.04	6.89 <sup>de</sup> ± 0.05
<b>40% crude protein</b>					
Diet III	355.31	75.70 <sup>a</sup> ± 0.06	65.80 <sup>a</sup> ± 0.49	19.59 <sup>d</sup> ± 0.05	7.18 <sup>abc</sup> ± 0.02
Diet IV	402.31	74.52 <sup>bc</sup> ± 0.09	61.36 <sup>d</sup> ± 0.05	23.18 <sup>c</sup> ± 0.05	6.76 <sup>e</sup> ± 0.02
<b>45% crude protein</b>					
Diet V	360.50	76.50 <sup>a</sup> ± 0.52	65.68 <sup>a</sup> ± 0.05	19.84 <sup>d</sup> ± 0.49	7.28 <sup>a</sup> ± 0.05
Diet VI	405.00	75.48 <sup>ab</sup> ± 0.60	62.12 <sup>c</sup> ± 0.02	25.40 <sup>a</sup> ± 0.06	7.05 <sup>bed</sup> ± 0.03
Control	406.50	73.97 <sup>c</sup> ± 0.03	58.00 <sup>e</sup> ± 0.14	22.62 <sup>c</sup> ± 0.22	7.01 <sup>cd</sup> ± 0.01

Results are mean ± SE of triplicate fish groups  
Values in each column with similar superscript are insignificantly different (p>0.05)

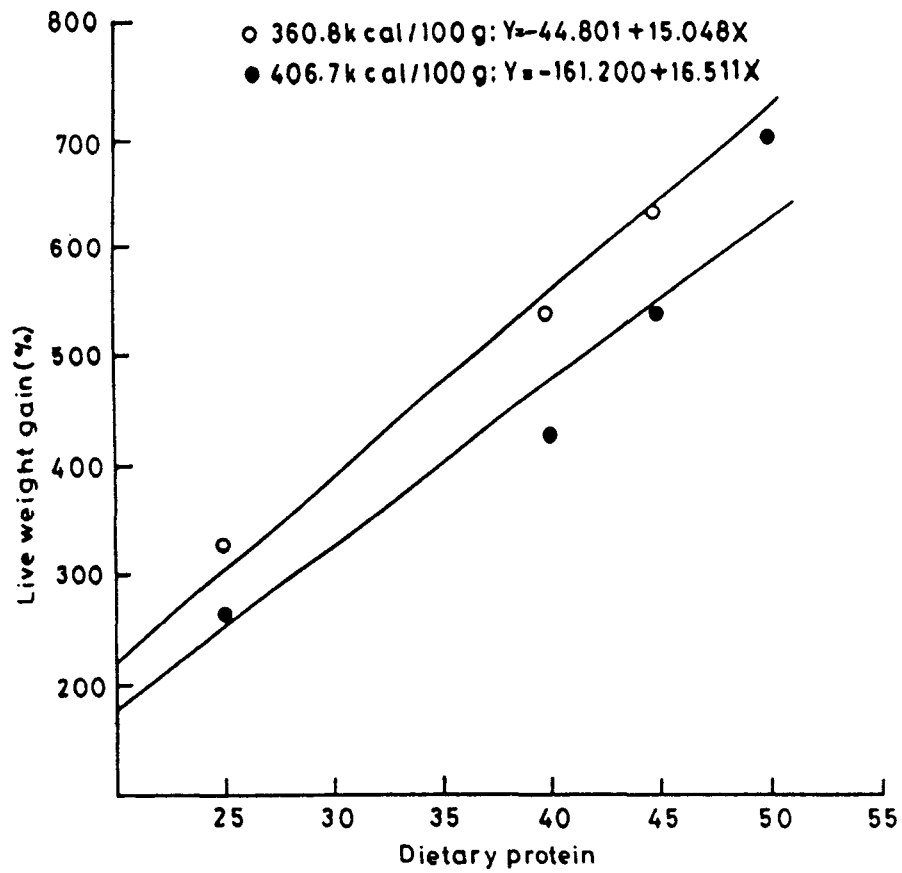


Fig. 1. Effect of dietary protein levels on live weight gain (%) of C. gariepinus fed experimental diets.



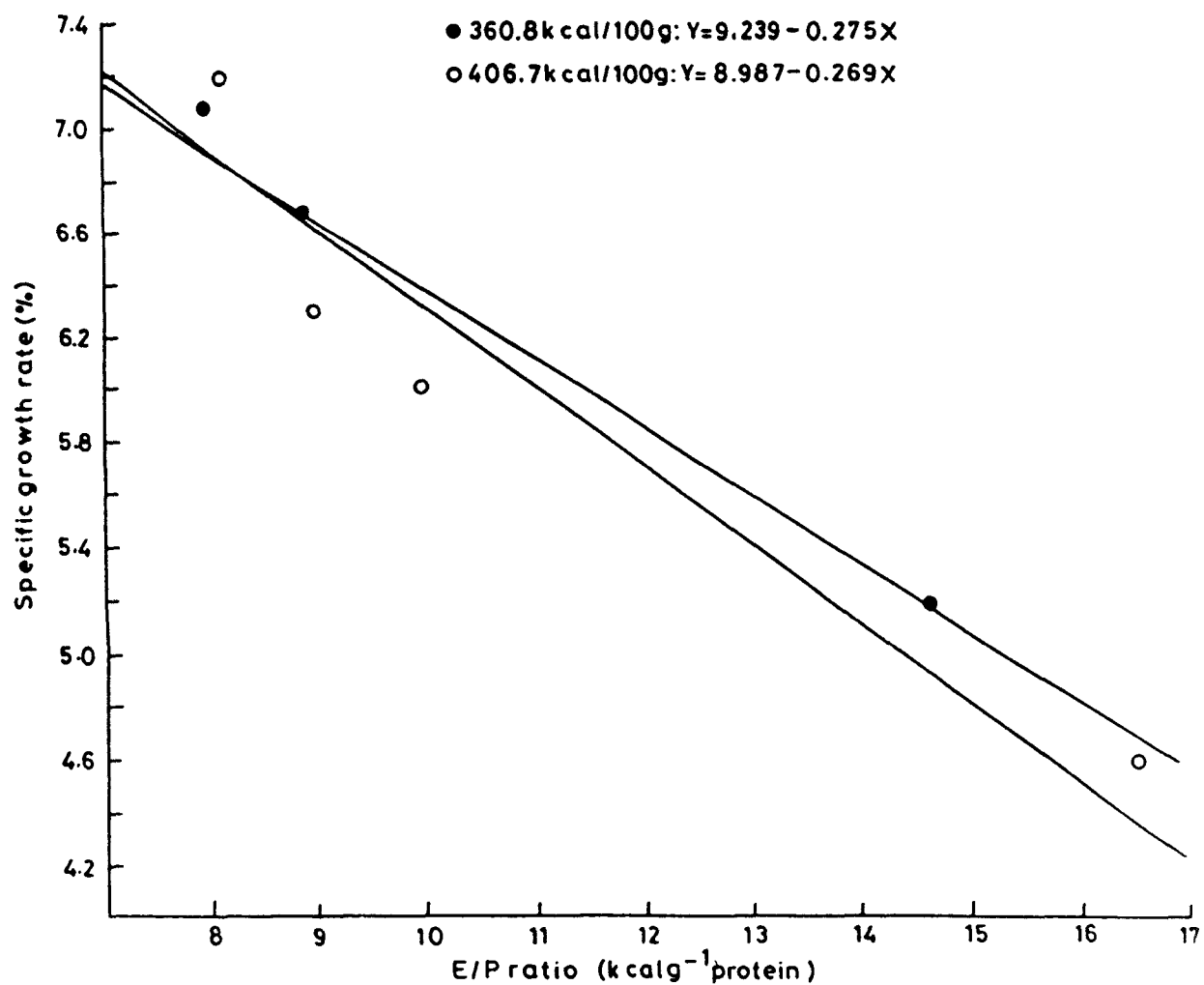


Fig. 2. Effect of dietary E/P ratios on specific growth rate (%) of C. gariepinus fed experimental diets.

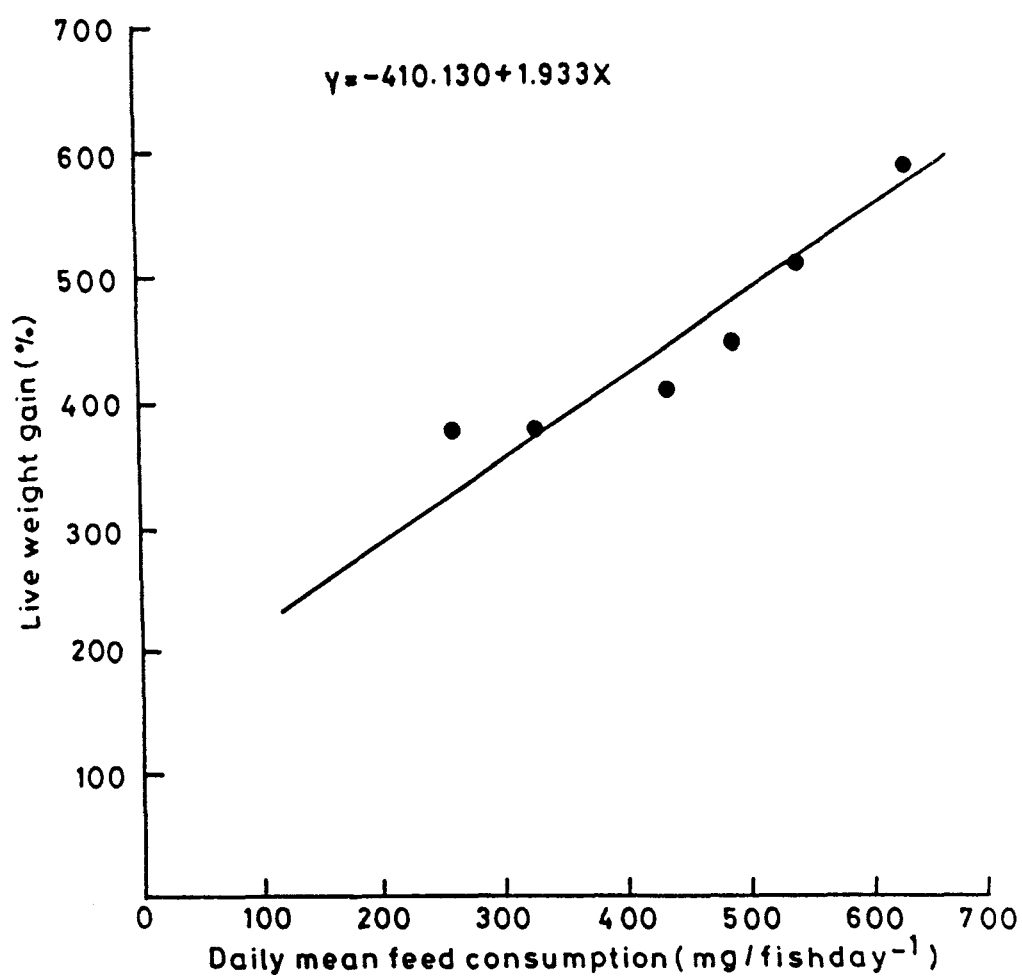


Fig. 3. Relationship between live weight gain (%) and daily mean feed consumed (mg/fish day<sup>-1</sup>) in C. gariepinus fed experimental diets.

## CHAPTER II

### OPTIMUM FEEDING RATE, ENERGY AND PROTEIN MAINTENANCE REQUIREMENTS OF *CLARIAS GARIEPINUS* (BURCHELL 1822)

#### INTRODUCTION

Optimization of feeding rate of cultured fish is important to achieve efficient production. Since feeding rate in fish affects their nutrient requirements, knowledge of optimal feeding rate is considered a prerequisite to nutrient requirement estimates (Tacon and Cowey, 1985; and Talbot, 1985).

Ration level in fish is also reported to influence fish growth, utilization efficiencies and chemical composition (Reddy and Katre, 1979; and Reinitz, 1983<sup>ab</sup>). Several factors, including fish size, feeding level and water temperature influence optimum feed requirements. In commercial culture, although control of environmental temperature may not be feasible, ration size can be manipulated to maximize production. Although some information is available on the nutrition of *C. gariepinus* (Henken *et al.*, 1985, 1987; Degani *et al.*, 1989; Uys, 1989; Mybenka and Agua, 1990; Hoffman and Prinsloo, 1995; Awaiss and Kestemont, 1998; and Murty and Naik, 1999), there is almost no information available on the optimal feeding rate of this species. Henken *et al.* (1985) have examined the effect of feeding level on apparent digestibility of this fish. A quantitative estimate of maximum daily feed intake of *C. gariepinus* fingerling has also been made by Hossain *et al.*<sup>b</sup> (1998).

The present study deals with the effect of feeding rate on growth and body composition of fingerling *C. gariepinus*, leading to the estimation of its optimal ration level, and energy and protein maintenance requirements. The information will be of interest to fish nutritionists and farmers.

## **MATERIALS AND METHODS**

### ***Preparation of experimental diet***

45% crude protein diet with 360.5 kcal /100g digestible energy (Table 1) was prepared using the method as described earlier under General Methodology section (page 6 ). Dietary energy was calculated using physiological fuel values, 3.5, 4.5 and 8.5 kcal/g for carbohydrate, protein and fat, respectively (Jauncey, 1982). An energy to protein ratio of 8.13 kcal/g was maintained in the diet as determined in the previous experiment (see Chapter I).

### ***Feeding trial***

Details of acclimation of fish and general experimental design have been described elsewhere. A 3 x 2 factorial design of experiment was used. Fish ( $6.14 \pm 0.13$  cm, total length; and  $1.70 \pm 0.01$ g weight) were stocked in triplicate groups of 15 fish each in circular polyvinyl troughs (water volume 55 L; water exchange rate 1L/min). Water temperature over the experimental period was  $25 \pm 1^\circ\text{C}$ . Fish were fed ration levels at 0-10% of body weight per day, on dry to wet weight basis, twice daily at 0800 and 1600h for 4 weeks. Mass weight of fish were taken weekly and ration size recalculated for subsequent feeding. No unconsumed feed was noticed in experimental troughs initially but in the last week of experiment fish took relatively longer time to consume

their daily ration and some unconsumed feed accumulated in higher ration groups.

### ***Estimation of gross energy and proximate analysis***

For initial and final carcass composition, fish were taken out from the acclimated stock and at the end of feeding trial from each trough and analysed for their proximate carcass composition, using standard techniques as given elsewhere (see page 7-9 ). Gross energy in the tissue was calculated using physiological fuel values similar to those used for the energy estimates in the diet.

### ***Statistical analysis***

Comparison among different treatment means were made by one way analysis of variance (Snedecor and Cochran, 1967) and Duncan's multiple range test (Duncan, 1955). A significance level of  $P < 0.05$  was used. Regression and correlation coefficient ( $r$ ) were calculated to establish the relationship between various parameters.

## **RESULTS**

Results of the 4 - week feeding trial have been summarized in Table 2- 4. A linear increase ( $r=0.99$ ) in average daily growth (Fig 1-2) was obtained by feeding fish with varying ration levels up to 8% Bw. day<sup>-1</sup>, corresponding to protein and energy intakes of 0.15g and 1.16 kcal energy/ fish day<sup>-1</sup>, respectively. In the linear growth portion, average daily growth increment of fish (Y) over increasing levels of protein and energy (X) was described by the equations  $Y=0.021+4.476 X$  for protein and  $Y=0.028+0.556 X$  for energy. Fish fed 10% Bw. day<sup>-1</sup> produced insignificant ( $P > 0.05$ ) difference in growth compared with those fed 8%

Bw. day<sup>-1</sup>. A continuous loss in weight was noticed in starved fish. A linear increase ( $r=0.95$ ) in specific growth rate (SGR%) was also evident with increase in ration levels up to 8% Bw. day<sup>-1</sup>(Fig 3).

Highest gains in body protein and energy were observed by feeding fish with 0.15g protein/fish day<sup>-1</sup> and 1.16 kcal/fish day<sup>-1</sup> but maximum gross growth efficiency, protein and energy conversion efficiencies were discernible at the ration level of 4% Bw. day<sup>-1</sup>, corresponding to protein and energy intakes of 0.05g and 0.39 kcal/fish day<sup>-1</sup>. When the same parameters were compared with those at 6% Bw. day<sup>-1</sup>, corresponding to 0.09g protein/ fish day<sup>-1</sup> and 0.73 kcal energy/fish day<sup>-1</sup>, no significant difference could be seen.

The best feed conversion ratio (FCR) was obtained in fish fed 4% Bw. day<sup>-1</sup> (0.05g protein/ fish day<sup>-1</sup> and 0.39 kcal energy/fish day<sup>-1</sup>). FCR was poor above and below this feeding level. FCR plotted against ration levels produced a typical U-shaped curve (Fig 4).

The proximate composition of fish fed varying ration levels has been given in Table 4. The body composition varied with levels of feeding. Changes in moisture, lipid and ash percentages were significant ( $P<0.05$ ), whereas variations in protein content were insignificant ( $P>0.05$ ). Moisture and ash decreased with increase in ration levels from 2 to 8% Bw. day<sup>-1</sup>. An increasing trend was also observed in body fat and energy (kcal g<sup>-1</sup>dry matter) contents. No significant difference in the proximate composition was observed between the fish fed 8% Bw. day<sup>-1</sup> and those fed 10% Bw. day<sup>-1</sup>. In starved fish, compared to crude protein, fat was greatly reduced, whereas moisture and ash contents registered an increase.

## DISCUSSION

It is apparent from the results of the present study that, over the experimental period, fingerling *C. gariepinus* fed ration at 2% Bw. day<sup>-1</sup> increased in its live weight by 0.11g/fish day<sup>-1</sup> whereas those receiving ration at 4% Bw. day<sup>-1</sup> increased by 0.29g/fish day<sup>-1</sup>. Although significant growth improvement was noticeable at higher ration levels, highest conversion values were achieved at a ration level of 4%, which was not significantly different from the values achieved when fish were fed at 6% Bw. day<sup>-1</sup>. This indicates that between a feeding rate of 4 to 6% Bw. day<sup>-1</sup>, a larger portion of dietary nutrients were utilized by fish for their growth.

Poor daily average growth increment and FCR in fish fed 2% Bw. day<sup>-1</sup> suggests that this ration level approximates only the maintenance requirement of nutrients, wherein a major portion of ingested nutrients is utilized to maintain life and a smaller portion available for growth. Hassan and Jafri (1994) obtained comparable results on the other *Clarias* species, *C. batrachus*. The findings also seem in agreement with the observations of Hung and Lutes (1987) on *Acipenser transmontanus* and Lupatsch et al. (1998) on *Sparus aurata*.

A positive effect of feeding rate on growth has been shown in striped bass (Hung et al., 1993) and in channel catfish (Li and Lovell, 1992). In these fishes, either a linear increase or plateauing effect was noted on the growth while feed conversion ratio reduced with increased ration. Similar pattern was observed in *C. gariepinus* during the present study. When FCR was plotted against ration level, a typical U-shaped curve was obtained, indicating that ration level of 4% Bw. day<sup>-1</sup> is optimum for the fingerling *C. gariepinus*. Similar results have been reported in other fish species (Hassan and Jafri, 1994; and Panda et al,

1999). Poor FCR at lower and higher ration levels can be the result of loss of nutrients and wastage of food, as fish took longer time to consume food to reach satiation (Tvenning and Giskegjerde, 1997). Although maximum growth improvement (g/fish day<sup>-1</sup>) was observed in *C. gariepinus* at a ration level of 8% Bw. day<sup>-1</sup>, which was not significantly different from the growth increment obtained when fish were fed at a ration level of 10% Bw. day<sup>-1</sup>, maximum gross growth efficiency was obtained when fish received ration at 4% Bw. day<sup>-1</sup>.

A gradual decline in conversion efficiency was noticed in fish fed at higher ration levels, thus feeding fish beyond a ration level of 4% Bw. day<sup>-1</sup> appears a wastage. A general reduction in conversion efficiency of fish at higher ration levels has also been reported by Hassan and Jafri (1994) in *C. batrachus*.

Several factors, including growth and diet, are known to influence the proximate composition of fish. Proximate composition of fish is also influenced by varying ration levels. In the present study, moisture, fat and ash contents of fish were significantly influenced by feeding rates. However, no significant change was observed in protein content. When fish were starved, amount of moisture and ash increased, whereas a distinct decrease in fat was noticeable over their initial values. Similar changes in proximate composition vis-à-vis ration level was noticed in *C. batrachus* (Hassan and Jafri, 1994). A net loss of energy in starving *C. gariepinus* indicates that both lipid and protein get catabolized during starvation, but there seems to be a preferential catabolism of body lipid. A slightly lower percentage of body fat was observed in fish fed lower ration levels, though at the same time the fish could manage to maintain relatively higher and constant amount of protein in their body tissue over the initial value, suggesting that in this fish body fat is preferred as energy source over protein. This finding finds support in the studies



made on other fishes (Hung and Lutes, 1987; Brown *et al.*, 1990; and Hassan and Jafri, 1994).

## SUMMARY

Optimum feeding rate, energy and protein maintenance requirements are reported in the fingerling *C. gariepinus*, fed purified diet (45% C.P.; 360.5 kcal/100g energy) at 0-10% Bw. day<sup>-1</sup>. A linear increase ( $r = 0.99$ ) was observed in daily average growth increment up to a ration level of 8% Bw. day<sup>-1</sup> but maximum conversion efficiencies were obtained at 4% Bw. day<sup>-1</sup>, corresponding to protein and energy intakes of 0.05g/fish day<sup>-1</sup> and 0.39 kcal /fish day<sup>-1</sup> respectively. The study indicates that a ration level of 4% Bw. day<sup>-1</sup> is optimum for this species at  $25 \pm 1^{\circ}\text{C}$ . Poor FCR and daily average growth increment obtained at a ration level of 2% Bw. day<sup>-1</sup> suggests that this level approximates the maintenance requirements of the fish. Body moisture, fat and ash contents were significantly ( $P < 0.05$ ) affected by ration levels whereas variations in protein were insignificant ( $P > 0.05$ ).

**Table 1. Ingredient and proximate composition of experimental diet**

<b>Ingredients</b>	<b>g/100g (as fed)</b>
Casein (Vitamin free; 84.6% C.P)*	40.17
Gelatin (87.0% C.P)*	12.84
Dextrin	25.71
$\alpha$ -Cellulose	7.28
Oil mix (2:1 corn and cod liver oil)	8.00
Vitamins mix .	1.00
Mineral mix .	3.00
Carboxymethyl cellulose	2.00
<b>Proximate composition% (calculated)</b>	
Crude protein	45.00
Crude fat	8.00
Carbohydrate	26.00
Energy (kcal/100g)	360.50
E/p ratio (kcal/g)	8.01

\*Loba Chemie, India

**Table 2. Growth of *C.gariepinus* fed varying levels of experimental diet**

% (Bw. day <sup>-1</sup> )	Feeding rate		Total diet fed (g/fish)	Initial average weight (g)	Final average weight (g)	Growth increment (g/fish day <sup>-1</sup> )	Gross growth efficiency (%)
	Protein (g/ fish day <sup>-1</sup> )	Energy kcal/g per fish day <sup>-1</sup>					
0.00	0.00	0.00	0.00	1.67 ±0.00	1.16 ±0.05	-0.06 <sup>e</sup> ±0.00	-
2.00	0.02	0.15	2.09	1.72 ±0.01	2.79 ±0.03	0.11 <sup>d</sup> ±0.00	107.87 <sup>b</sup> ±3.68
4.00	0.05	0.39	5.38	1.72 ±0.01	4.85 ±0.03	0.29 <sup>c</sup> ±0.00	122.00 <sup>a</sup> ±0.85
6.00	0.09	0.73	10.07	1.73 ±0.00	7.46 ±0.01	0.51 <sup>b</sup> ±0.00	118.97 <sup>a</sup> ±0.43
8.00	0.15	1.16	16.09	1.70 ±0.01	9.90 ±0.17	0.73 <sup>a</sup> ±0.01	106.84 <sup>b</sup> ±0.90
10.00	0.18	1.46	20.32	1.69 ±0.01	9.82 ±0.14	0.74 <sup>a</sup> ±0.01	83.84 <sup>c</sup> ±0.89

Results are mean ± SE of triplicate fish groups

Values in each column with similar superscript are insignificantly different (P>0.05)

**Table 3. Conversion efficiencies in *C. gariepinus* fed varying levels of experimental diet**

B.W. day <sup>-1</sup>	Feeding Rate		Conversion efficiency (%)	
	Protein (g/fish day <sup>-1</sup> )	Energy (kcal/g per fish day <sup>-1</sup> )	Energy	Protein
0.00	0.00	0.00	-	-
2.00	0.02	0.15	13.80 <sup>c</sup> ±0.67	17.33 <sup>c</sup> ±0.51
4.00	0.05	0.39	18.79 <sup>a</sup> ±0.37	21.86 <sup>a</sup> ±0.09
6.00	0.09	0.73	18.69 <sup>a</sup> ±0.31	21.16 <sup>a</sup> ±0.10
8.00	0.15	1.16	17.19 <sup>b</sup> ±0.14	19.16 <sup>b</sup> ±0.32
10.00	0.18	1.46	13.50 <sup>c</sup> ±0.02	14.98 <sup>d</sup> ±0.07

Values are mean ±SE of triplicate fish groups

Means in each column with similar superscript are insignificantly different (P>0.05)

**Table 4. Proximate composition and energy content in the carcass of *C. gariepinus* fed varying levels of experimental diet**

Feeding rate			Proximate composition (g/100g dry matter)				
% (Bw. day <sup>-1</sup> )	Protein (g/fish day <sup>-1</sup> )	Energy (kcal/gper fish day <sup>-1</sup> )	Moisture (g/100g, wet weight)	Crude protein	Crude fat	Ash	kcal g <sup>-1</sup> dry matter
0.00	0.00	0.00	80.12 <sup>a</sup> ±0.13	79.04 <sup>a</sup> ±0.43	10.00 <sup>e</sup> ±0.58	8.93 <sup>a</sup> ±0.11	0.87 <sup>e</sup> ±0.01
2.00	0.02	0.15	78.93 <sup>b</sup> ±0.11	69.72 <sup>a</sup> ±0.44	12.01 <sup>d</sup> ±0.12	8.60 <sup>ab</sup> ±0.12	0.95 <sup>d</sup> ±0.01
4.00	0.05	0.39	77.01 <sup>c</sup> ±0.32	69.68 <sup>a</sup> ±0.42	16.00 <sup>c</sup> ±0.58	8.25 <sup>bc</sup> ±0.14	1.09 <sup>c</sup> ±0.02
6.00	0.09	0.73	76.68 <sup>c</sup> ±0.15	69.48 <sup>a</sup> ±0.39	18.50 <sup>b</sup> ±0.29	7.92 <sup>c</sup> ±0.05	1.13 <sup>b</sup> ±0.01
8.00	0.15	1.16	76.23 <sup>c</sup> ±0.01	69.32 <sup>a</sup> ±0.44	20.03 <sup>a</sup> ±0.01	7.75 <sup>c</sup> ±0.15	1.17 <sup>a</sup> ±0.00
10.00	0.18	1.46	76.25 <sup>c</sup> ±0.13	69.16 <sup>a</sup> ±0.07	20.00 <sup>a</sup> ±0.13	7.75 <sup>c</sup> ±0.26	1.17 <sup>a</sup> ±0.01
Initial	-	-	79.02 ±0.28	68.72 ±0.18	11.00 ±0.10	8.57 ±0.21	0.94 ±0.01

Values are mean ±SE of triplicate fish groups

Means in each column with similar superscript are insignificantly different (P>0.05)

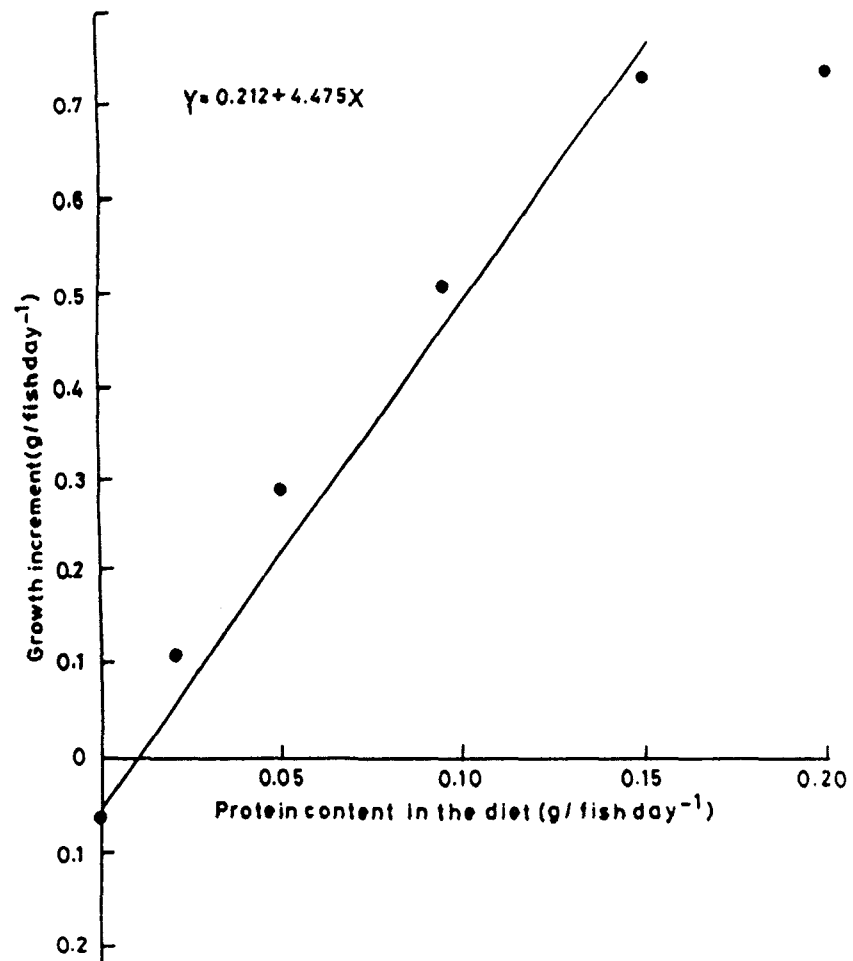


Fig. 1. Growth increment in *C. gariepinus* fed varying levels of dietary protein (g/lfish day<sup>-1</sup>).

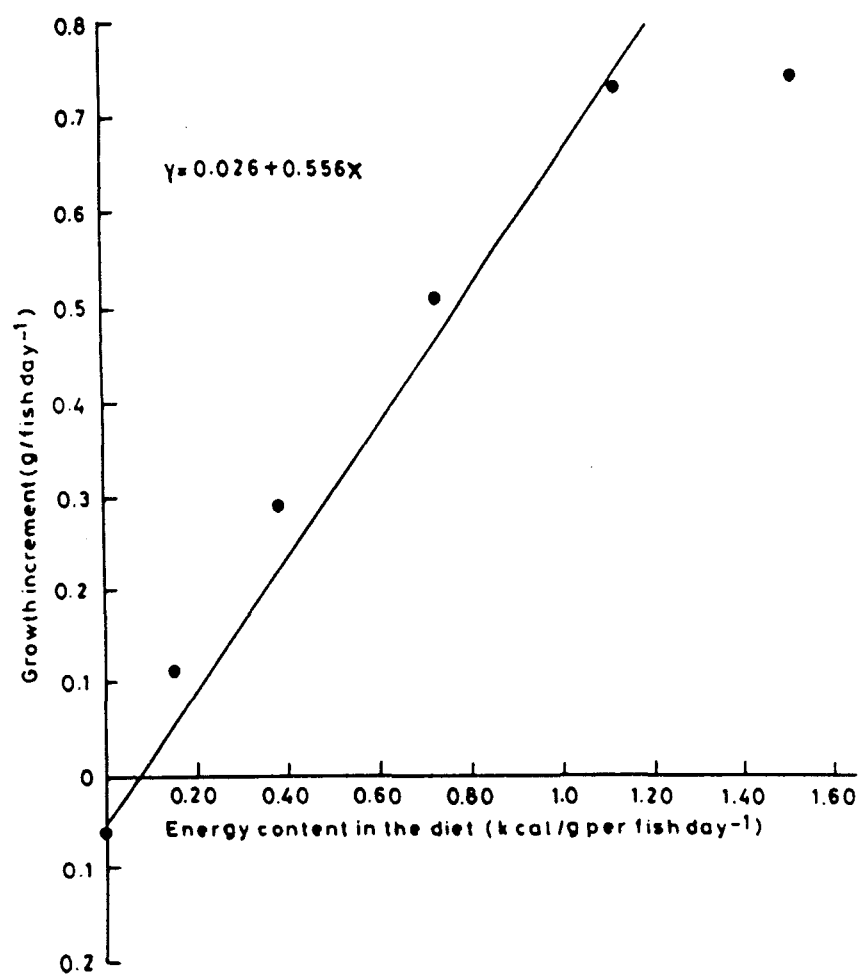


Fig. 2. Growth increment (g/fish day<sup>-1</sup>) in *C. gariepinus* fed varying levels of dietary energy (k cal/g per fish day<sup>-1</sup>).

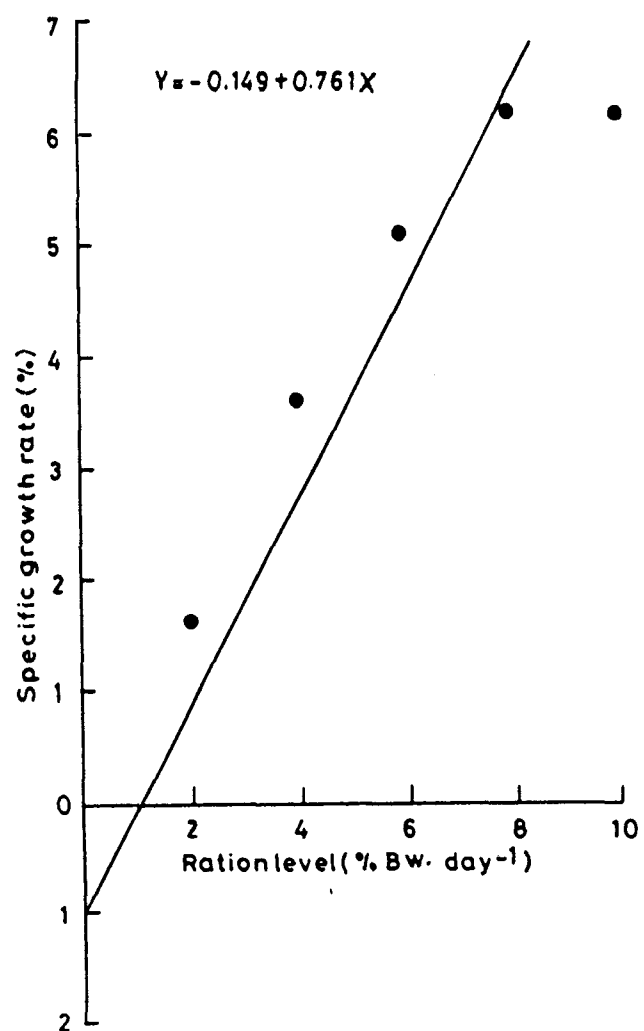


Fig. 3. Specific growth rate (%) in C. gariepinus fed varying ration levels.



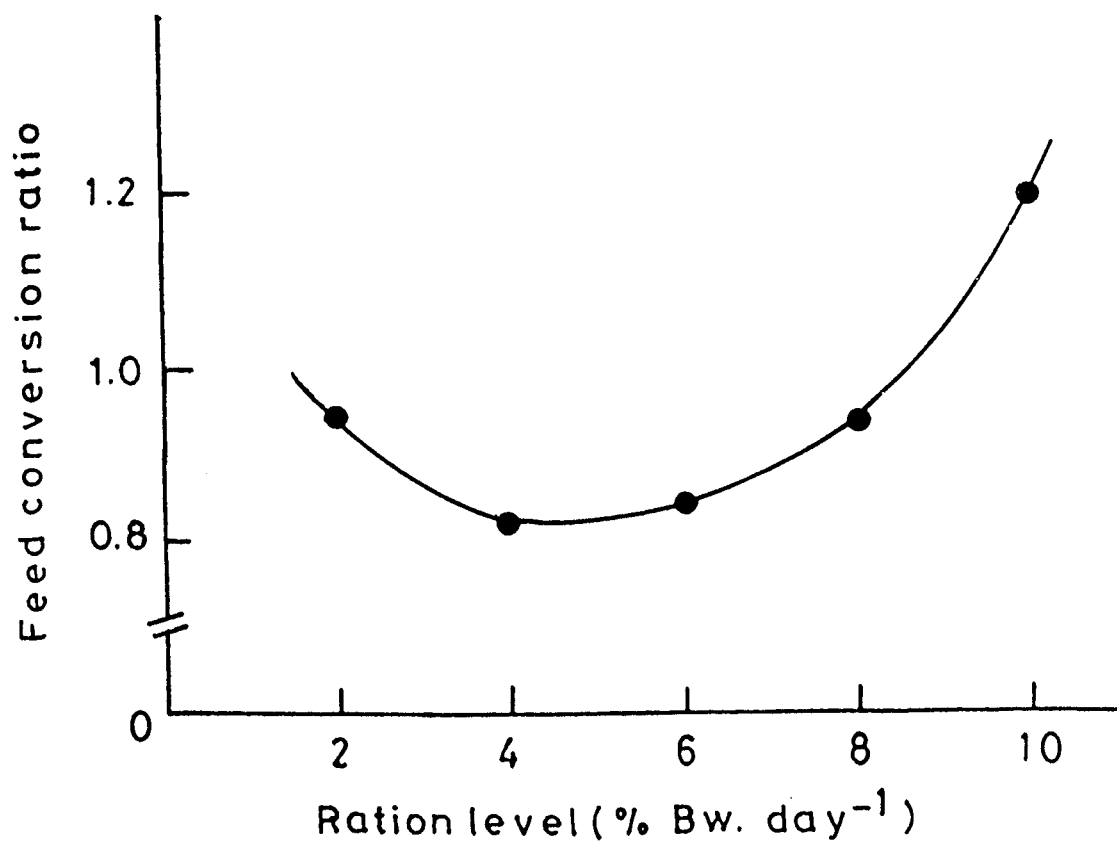


Fig. 4. Effect of ration levels (% B.W. day<sup>-1</sup>) on feed conversion ratio in C. gariepinus.

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